

Phantoms for evaluating the impact of skin pigmentation on photoacoustic imaging and oximetry performance: supplement

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Supplement 1

Blood Oxygenation Stability for Different Tubing Materials

Preliminary testing suggested that dithionite-treated blood volumes injected into tubes within PVCP phantoms did not have stable SO_2 . This was determined to be caused by oxygen gas permeation through the tube wall and phantom medium. To identify a tubing material that improved blood SO_2 stability, we performed PAI of a series of tubes suspended in a water bath, each made of a different material and filled with bovine blood desaturated to $\sim 50\%$ SO_2 (Fig. S1). We found that K-flex tubing offered high stability ($<2\%$ drift over 20 minutes), which may be due to its low oxygen gas permeability. This was similar to stability observed in glass capillary tubing, which is less suitable for PAI due to high acoustic impedance mismatch. We performed PAI of an array of water-filled K-flex tubes immersed in a water bath at depths of 5, 10, 15, 20, and 25 mm, acquiring five scans from 700-900 nm in 2 nm steps. Results verified that K-flex tubing contributes minimal PA signal without spectral variations (Fig. S2). Tube signals were $< 4 \times 10^4$ units, or $< 6\%$ of the amplitude for the lowest nigrosin concentration in Fig. 4.

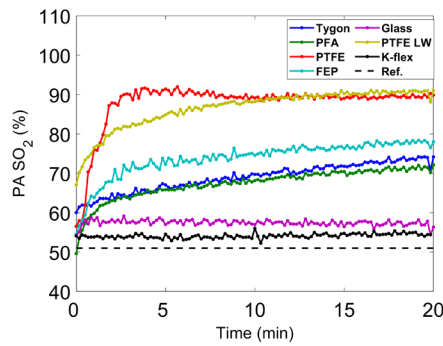


Fig. S1. Photoacoustic SO_2 measurements of deoxygenated blood over time in tubes of different material types in a water bath. K-flex tubing was found to achieve superior temporal stability similar to glass tubing, but without strong acoustic artifacts due to impedance mismatch between glass and water. (PFA: perfluoroalkoxy polymer; PTFE: polytetrafluoroethylene; FEP: fluorinated ethylene propylene; PTFE LW: polytetrafluoroethylene, light wall; K-flex: Kynar-flex.)

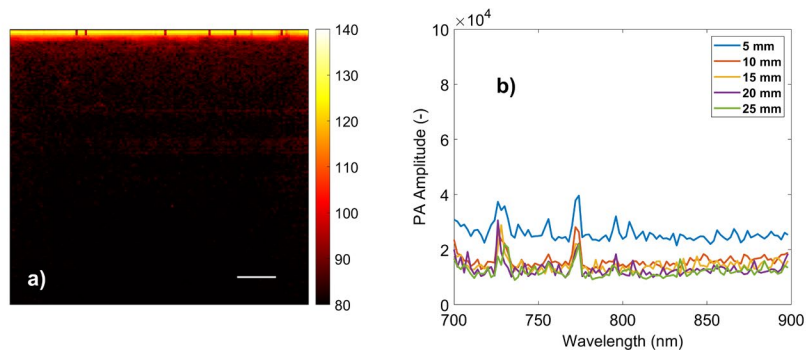


Fig. S2.a) PAI at 800 nm of K-flex tubes in a water bath. B) target photoacoustic amplitude spectra measured by ROI analysis.

Epidermis Thickness Measurements by OCT

A spectral-domain OCT system with $6 \mu\text{m}$ axial resolution, $25 \mu\text{m}$ lateral resolution was used to estimate thickness of epidermis-mimicking PVCP layers. To maximize OCT image contrast

of the epidermal layer, a PVCP skin layer phantom was prepared with a highly light scattering epidermis doped with 5 mg/mL TiO_2 and a dermis layer with no TiO_2 (Fig. S3). A water droplet was placed on the sample surface to reduce specular reflections. The sample was scanned over a 4 mm x 4 mm area (500 B-scans). For each A-scan (column), thickness was estimated as the distance between the top and bottom edges of the epidermis layer, defined as the first and last data points exceeding 50 grayscale units. Axial distance per pixel was calculated assuming a refractive index of 1.5 for PVCP. Thickness was averaged over 350 x 500 A-scans.

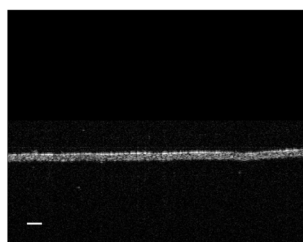


Fig. S3. Representative OCT image of PVCP skin-mimicking layer with turbid epidermis doped with TiO_2 (visible in image) and transparent dermis layer (not visible).

Clutter Removal from PA Oximetry Maps

Segmentation and removal of clutter from SO_2 maps is an outstanding challenge that may require complex algorithms. Unlike some clinical scenarios, phantom target locations are known *a priori*; we thus also generated SO_2 maps to illustrate best-case clutter reduction by only displaying SO_2 within 1.5 x 2 mm regions of interest (ROIs) drawn on the tubes (Fig S4).

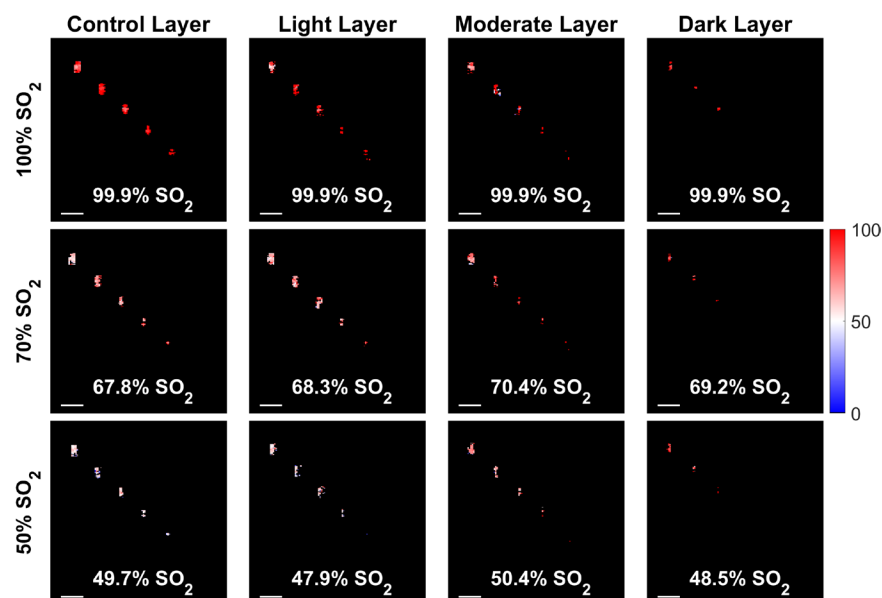


Fig. S4. SO_2 maps from Figure 8 with additional ROI thresholding to remove clutter.